Remarks

Claims 1-16, 20-29 and 48-59 are pending in this application. Claims 20-29 and 51-53 were previously withdrawn from consideration.

No new matter is added. Applicants respectfully request reconsideration of the application in view of the following remarks.

Claim Rejections Under 35 U.S.C. § 103(a)

Claims 1-16, 48-50 and 54-59 are rejected under 35 U.S.C. § 103(a) as allegedly being obvious over Vasa et al., (Circ. Res. 89(1):E1-7, 2001, "A"), Vasa et al. (Circulation 103(24): 2885-90, 2001, "B") and Scott et al. (Circulation 104: 491-496, 2001). Applicants respectfully disagree with this rejection.

I. The Cited References

Vasa *et al.* [A] disclose that the number of endothelial progenitor cells (EPCs) is reduced in subjects with coronary artery disease (CAD) as compared to subjects with no evidence of CAD by history or physical exam. In addition, EPCs from subjects with CAD had an impaired migratory response. Vasa *et al.* [A] suggest that the reduced number and function of EPCs correlates with adult neovascularization. Additionally, an analysis of various risk factors showed that smoking and age correlated with reduced numbers of EPCs. Vasa *et al.* [A] does not describe any subpopulations or confirm any associations of EPC with risk in the healthy control subjects. The control population of healthy subjects is identified as simply a single unified population, and no comparison is made between healthy subjects with different Framingham risk scores. Thus, Vasa *et al.* [A] does not suggest that a decrease in the number of EPCs will affect vascular function in a healthy subject without coronary artery disease, nor does it provide methods to identify those completely asymptomatic subjects without cardiovascular disease who are at risk for developing the disease.

Vasa *et al.*[B] describe that statin treatment of patients with symptomatic cardiovascular disease is associated with an increase in the number of circulating endothelial progenitor cells, as well as an increase in the migratory capacity in response to vascular endothelial growth factor of

endothelial progenitor cells. Additionally, Vasa *et al.*[B] discloses that healthy subjects treated with Atorvastatin experienced an increase in circulating EPC numbers compared to healthy control subjects. Thus, Vasa *et al.*[B] suggest that the increase in the number and migratory activity of EPCs resulting from statin treatment contributes to the angiogenesis and vasculogenesis that lead to neovascularization in subjects with coronary artery disease. However, Vasa *et al.*[B] do not teach that an increase in EPC number *in healthy subjects without symptomatic cardiovascular disease* is associated with any kind of increased vascular function. Further, the teachings of this reference are directed towards the treatment of subjects with cardiovascular disease with Atorvastatin, does not provide methods for detecting vascular function. Vasa *et al.*[B] does teach that there is an increased number of circulating EPCs in healthy subjects treated with Atorvastatin, but does not suggest, nor render obvious, that this increase is associated with increased vascular function *in healthy subjects without symptomatic cardiovascular disease,* and thus does not provide any indication of which asymptomatic subjects have reduced risk for developing cardiovascular disease in the future.

Grundy et al. (Circulation 104: 491-496, 2001, termed "Scott et al." in the Office action), teach the use of Framingham Heart Study (FHS) , and discloses that in order to be widely useful, it must be determined if the FHS results estimates are of use widely in the U.S. population (see page 491, first column). Grundy et al. describe studies that were performed to "evaluate the transportability of FHS risk equations" (see page 492), such as in subjects with new-onset coronary heart disease (CHD) in a number of populations, such as African Americans, older adults, Native Americans, Asian Americans, Hispanic Americans, "white" populations, women, young adults, and people with a high socioeconomic status. Grundy et al. identify a number of research needs, including improving the predictive power of the FHS (see page 495, second column, first bullet point), and evaluating risk assessment for predictions of risk in patients with established cardiovascular disease (CVD, see page 495, second column, 4th bullet point). Given the large amount of research indicated by Grundy to be required, it is difficult to determine why this publication can be seen to provide a reasonable expectation that any method would be effective that utilized the FHS results, let alone the claimed methods.

II. Unpredictability in the Clinical Arts

The Office action seems to allege that since Vasa *et al.*[A] and Vasa *et al.*[B], demonstrate that EPCs are reduced in patients with coronary artery disease, and that patients with multiple risk factors have lower EPCs than patient with CAD (but with less risk factors), then it would be obvious that EPCs would be lower in healthy subjects. The Office action uses Grundy et al. simply to show that Framingham Risk Scores were well known.

The Office action asserts that all the elements of the claimed method were known in the art. However, this is incorrect: the prior art simply does not teach that the number of endothelial cells can be used to determine risk for any disease in asymptomatic subjects. Thus, all of the elements of the claimed method are not disclosed in the prior art.

The Office action further argues that there is a reasonable expectation of success that the claimed methods world be successful in view of the cited prior art. As set forth in MPEP § 2143. 02, to support a finding of obviousness, it must be demonstrated that the claimed combination would yield nothing more than predictable results to one of ordinary skill in the art. Applicants submit that the effectiveness of the claimed methods simply could not be predicted based on the cited prior art.

Clinical markers of advanced disease, or of disease progression, cannot necessarily be used to determine which asymptomatic individuals are at risk for developing a disease in the future. The lack of predictability of a marker (such as a type of cell) for predicting risk for the development of a disease, even when that marker is proven to be associated with frank disease, is rampant in clinical science. Two examples are provided below in the fields of AIDS research and cancer research. These examples document the unpredictability of markers for identifying a subject with increased disease risk.

CD4 count is used to define AIDS in subjects infected with HIV. De Wolf et al. (AIDS 11(15): 1799-1806, 1997, abstract attached) studied HIV-1 RNA, CD4+ T cell count, and T cell function in subjects with AIDS. At one year of seroconversion, the HIV-1 RNA level was the only marker that was strongly predictive of progression to AIDs. Low CD4+ T cell count was

only predictive for progression to AIDS 2-5 years after seroconversion. Thus, early after infection, CD4+ T cell count was not predictive as to which subjects would develop AIDS. This documents that, at the time the present application was filed, one of skill in the art would not predict that a marker, such as the amount of specific type of cell, that could be used to detect advanced disease, even if that type of cell is of use late in the disease process to ascertain disease progression.

It should be noted that there are later publications that provide validation of the conclusions of de Wolf et al.. For example, Badri et al. (BMC Infectious Diseases 8(89): 1-8, 2008) describes the utility of CD4 counts for early prediction of virological failure during antiretroviral therapy. CD4 counts correlated with viral load, but had "very limited utility in identifying virological failure in individual patients....the association between CD4 count slope and virological failure was poor" (see the abstract).

Another example documenting that a marker which is of use late in a disease process cannot be used to identify those asymptomatic individuals who will develop the disease is found in the field of cancer biology. Carcinoembryonic Antigen (CEA) is a glycoprotein that is produced in significant amounts by the large intestine during fetal development, and it is expressed in colon adenocarcinoma, but not in normal bowel. CEA is a useful marker for determining the prognosis of colon cancer, as it is expressed in advanced disease. High CEA levels can be used to stage colon cancer. However, CEA expression is not a good early diagnostic test for colon cancer (see "Carcinoembryonic Antigen (CEA), **http://www.patient.co.uk.showdoc/40025955/**, copy enclosed). This documents that a marker that can be successfully used in diseased individuals (those with colon cancer) cannot be used to determine which asymptomatic individuals (healthy subjects) will develop the disease (colon cancer).

This evidence discussed above documents that one of skill in the art would not expect that a marker that is of use late in a disease process could predictably be used to identify asymptomatic individuals with the disease, or could be used to predict which individuals would develop the disease. A *prima facie* case of obviousness simply is not supported by the cited

references, given the unpredictability in the art.

III. Evidence that Teaches Away

There is another striking example in the field of heart disease. Ventricular arrhythmias provide prognostic value for survival in subjects with significant coronary artery disease. However, there was no relationship between ventricular arrhythmia and survival in subjects without significant coronary artery disease (Clairff et al., J. American College Cardiol. 1983: 1060-67, see attached abstract). Similarly, Fleg et al. (Am. J. Cardiol. 54: 762-764, 1984) disclose that exercised-induced ventricular tachycardia is associated with significant heart disease. However, exercised-induced ventricular tachycardia in apparently healthy subjects does not predict any increased cardiovascular morbidity or mortality (Fleg et al., Am. J. Cardiol. 54: 762-764, 1984). Thus, one of skill in the art would predict that a test that is of use for determining an outcome in subject with coronary artery disease would not be predictive for the outcome in asymptomatic individuals. The scientific evidence in the prior art teaches away from the arbitrary conclusions asserted in the Office action.

Based on the scientific evidence, one of skill in the art would understand that tests that predict risk in subjects with coronary artery disease could not be used to identify decreased vascular function in a subject without symptomatic cardiac disease. Based on the teachings of Fleg et al. and Clairff et al., that one of skill in the art would predict that any test disclosed in Vasa et al.[A] or Vasa et al.[B] would simply not be of use in a subject that does not have symptomatic cardiovascular disease.

IV. Missing Elements: Claims 48-50

In addition, with regard to claims 48-50, Vasa *et al.* [A] do not teach assaying a number of senescent endothelial progenitor cells in a blood sample from a subject. The Office action equates age with senescence (see page 3). This is simply incorrect.

As discussed in the prior response, the age of a subject is not synonymous with the number of senescent EPCs in blood samples taken from that subject. The US PTO has simply

not provided any scientific evidence that age and the number of senescent endothelial cells are correlated, but has only provided conclusory statements. This is not the proper standard. MPEP § 2141 states:

> "The Supreme Court in *KSR* noted that the analysis supporting a rejection under 35 U.S.C. **103** should be made explicit. The Court quoting *In re Kahn,* 441 F.3d 977, 988, 78 USPQ2d 1329, 1336 (Fed. Cir. 2006), stated that "'[R]ejections on obviousness cannot be sustained by mere conclusory statements; instead, there must be some articulated reasoning with some rational underpinning to support the legal conclusion of obviousness.'" *KSR*, 550 U.S. at ___, 82 USPQ2d at 1396."

Indeed, in the present instance, not only is the US PTO relying on conclusory statements, these conclusory statements are completely contrary to the evidence of record. Specifically, Erusalimsky and Kurz (Handbook of Experimental Pharmacology 176: 213-248, 2006, copy included with the prior response), provided evidence that endothelial cell senescence can be induced by genetic discorders, teleomere damage, oxidative stress and sustained mitogenic stimulation, as well as age. Eursalimky and Kurz indicate that there is data that "argues for and against a role of endothelial cell senescence in age-related vascular pathology." Thus, it simply cannot be inferred that age will be simply correlated with the number of senescent EPCs, especially if the subjects are exposed to oxidative stress (such as smoking).

None of the prior art discloses assaying a number of senescent endothelial progenitor cells in a blood sample from the subject, wherein a senescent endothelial progenitor cell is a viable endothelial cell that exhibits clonal exhaustion. In addition, even if, for some imperceptible reason, one of skill in the art equated age with senescence, this does not negate the unpredictability in the art. As documented above, it cannot be predicted that a marker that is of use late in a disease process can be used to identify asymptomatic individuals with the disease, or can be used to predict which individuals would develop the disease. Thus, with regard to claims 48 -50, maintaining the present obviousness rejection is improper.

V. No Reasonable Expectation of Success

To support an obviousness rejection, one of skill in the art must reasonably expect the claimed combination to work (see In re O'Farrell, 853 F.2d 894, 903-904, 7 USPQ2d 1673, 1681 (Fed Cir. 1988)). M.P.E.P. § 2143 documents that in order to support a *prima facie* case of obviousness, it must be documented that the claimed methods would be predictable to one of skill in the art based on the prior art. In the present case, the cited prior art simply does not provide any evidence of the predictability of the claimed methods.

This evidence presented herein demonstrates that even if the number of endothelial cells can be used to predict neovascularation, it clearly cannot be predicted that the number of endothelial cells could be used to identify a subject with increased cardiovascular risk or decreased vascular function, wherein a decrease in the number of endothelial progenitor cells in the sample as compared to the control indicates decreased vascular function in the subject.

Reconsideration and withdrawal of the rejection are respectfully requested.

Conclusion

Applicants believe that this application is in condition for allowance, which action is requested. If any issues remain, or if the rejection under 35 U.S.C. § 103 is maintained, the Examiner is formally requested to contact the undersigned prior to issuance of the next Office Action in order to arrange a telephonic interview. It is believed that a brief discussion of the merits of the present application may expedite prosecution. This request is being submitted under MPEP § 713.01, which indicates that an interview may be arranged in advance by a written request.

Respectfully submitted,

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